CLAIMS

1. A polypeptide comprising an immunogenic portion of a native WT1, or a variant thereof that differs in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with WT1-specific antisera and/or T-cell lines or clones is not substantially diminished, wherein the polypeptide comprises no more than 16 consecutive amino acid residues present within a native WT1 polypeptide.

2. A polypeptide according to claim 1, wherein the immunogenic portion binds to an MHC class I molecule.

3. A polypeptide according to claim 1, wherein the immunogenic portion binds to an MHC class II molecule.

4. A polypeptide according to claim 1, wherein the polypeptide comprises a sequence selected from the group consisting of:

(a) sequences recited in one or more of Tables II - XLVI;

(b) variants of the foregoing sequences that differ in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with antigen-specific antisera and/or T-cell lines or clones is not substantially diminished; and

(c) mimetics of the foregoing sequences, wherein the ability of the mimetic to react with antigen-specific antisera and/or T-cell lines or clones is not substantially diminished.

5. A polypeptide according to claim 1, wherein the polypeptide comprises a sequence selected from the group consisting of:

(a) ALLPAVPSL (SEQ ID NO:34), GATLKGVAA (SEQ ID NO:88), CMTWNQMNL (SEQ ID NOs: 49 and 258), SCLESQPTI (SEQ ID NOs: 199 and 296),

SCLESQPAI (SEQ ID NO:198), NLYQMTSQL (SEQ ID NOs: 147 and 284); ALLPAVSSL (SEQ ID NOs: 35 and 255), RMFPNAPYL (SEQ ID NOs: 185 and 293);

(b) variants of the foregoing sequences that differ in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with antigen-specific antisera and/or T-cell lines or clones is not substantially diminished; and

(c) mimetics of the foregoing sequences, wherein the ability of the mimetic to react with antigen-specific antisera and/or T-cell lines or clones is not substantially diminished.

6. A polypeptide according to claim 1, wherein the polypeptide comprises 4-16 consecutive amino acids of a native WT1 polypeptide.

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7. A polypeptide according to claim 1, wherein the polypeptide comprises 8-10 consecutive amino acids of a native WT1 polypeptide.

8. A polypeptide comprising an immunogenic portion of amino acid residues 1 - 174 of a native WT1 polypeptide, or a variant thereof that differs in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with WT1-specific T-cell lines or clones is not substantially diminished, wherein the polypeptide comprises no more than 16 consecutive amino acid residues present within amino acids 175 to 449 of the native WT1 polypeptide.

9. A polypeptide comprising a variant of an immunogenic portion of WT1 that differs in substitutions at between 1 and 3 amino acid positions within the immunogenic portion, such that the ability of the variant to react with WT1-specific antisera and/or T-cell lines or clones is enhanced relative to a native WT1.

10. A mimetic of an immunogenic portion of a WT1 polypeptide, wherein at least one amino acid residue is replaced by a compound that is not an amino acid, such that the

ability of the mimetic to react with antigen-specific antisera and/or T-cell lines or clones is not diminished.

11. A polynucleotide encoding a polypeptide according to claim 1 or claim 8.

12. A phamaceutical composition, comprising:

(a) a polynucleotide encoding a WT1 polypeptide, wherein the polypeptide comprises an immunogenic portion of a native WT1 or a variant thereof that differs in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with antigen-specific antibodies and/or T cell lines or clones is not substantially diminished; and

(b) a pharmaceutically acceptable carrier or excipient.

13. A pharmaceutical composition, comprising:

(a) an antibody or antigen-binding fragment thereof that specifically binds to a WT1 polypeptide; and

(b) a pharmaceutically acceptable carrier or excipient.

14. A pharmaceutical composition, comprising:

(a) a T cell that specifically reacts with a WT1 polypeptide; and

(b) a pharmaceutically acceptable carrier or excipient.

15. A pharmaceutical composition, comprising:

(a) an antigen presenting cell that expresses

(i) a WT1 polypeptide that comprises an immunogenic portion of a native WT1 or a variant thereof that differs in one or more substitutions, deletions, additions

and/or insertions such that the ability of the variant to react with antigen-specific antibodies and/or T cell lines or clones is not substantially diminished; and

(b) a pharmaceutically acceptable carrier or excipient.

16. A method for enhancing or inducing an immune response in a human patient, comprising administering to a patient a pharmaceutical composition comprising:

(a) a WT1 polypeptide that comprises an immunogenic portion of a native WT1 or a variant thereof that differs in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with antigen-specific antibodies and/or T cell lines or clones is not substantially diminished; and

(b) a physiologically acceptable carrier or excipient;

and thereby enhancing or inducing an immune response specific for WT1 or a cell expressing WT1 in the human patient.

17. A method for enhancing or inducing an immune response in a patient, comprising administering to a patient a pharmaceutical composition according to any one of claims 12-15.

18. A method for enhancing or inducing an immune response in a human patient, comprising administering to a patient a vaccine comprising:

(a) a WT1 polypeptide that comprises an immunogenic portion of a native WT1 or a variant thereof that differs in one or more substitutions, deletions, additions and/or insertions such that the ability of the variant to react with antigen-specific antibodies and/or T cell lines or clones is not substantially diminished; and

(b) a non-specific immune response enhancer;

and thereby enhancing or inducing an immune response specific for WT1 or a cell expressing WT1 in the human patient.

19. A method for stimulating and/or expanding T cells, comprising contacting T cells with a WT1 polypeptide, a polynucleotide encoding a WT1 polypeptide and/or an antigen presenting cell that expresses a WT1 polypeptide under conditions and for a time sufficient to permit the stimulation and/or expansion of T cells.

20. A method according to claim 19, wherein the T cells are present within bone marrow, peripheral blood or a fraction of bone marrow or peripheral blood.

21. A method according to claim 19, wherein the bone marrow, peripheral blood or fraction is obtained from a patient afflicted with a malignant disease associated with WT1 expression.

22. A method according to claim 19, wherein the bone marrow, peripheral blood or fraction is obtained from a mammal that is not afflicted with a malignant disease associated with WT1 expression.

23. A method according to claim 19, wherein the T cells are cloned prior to expansion.

24. A method for stimulating and/or expanding T cells in a mammal, comprising administering to a mammal a pharmaceutical composition comprising:

(a) one or more of:

(i) a WT1 polypeptide;

(ii) a polynucleotide encoding a WT1 polypeptide; or

(iii) an antigen-presenting cell that expresses a WT1 polypeptide; and

(b) a physiologically acceptable carrier or excipient;

and thereby stimulating and/or expanding T cells in a mammal.

25. A method for monitoring the effectiveness of an immunization or therapy for a malignant disease associated with WT1 expression in a patient, comprising the steps of:

(a) incubating a first biological sample with one or more of:

(i) a WT1 polypeptide;

(ii) a polynucleotide encoding a WT1 polypeptide; or

(iii) an antigen-presenting cell that expresses a WT1 polypeptide

wherein the first biological sample is obtained from a patient prior to a therapy or immunization, and wherein the incubation is performed under conditions and for a time sufficient to allow immunocomplexes to form;

(b) detecting immunocomplexes formed between the WT1 polypeptide and antibodies in the biological sample that specifically bind to the WT1 polypeptide;

(c) repeating steps (a) and (b) using a second biological sample obtained from the patient following therapy or immunization; and

(d) comparing the number of immunocomplexes detected in the first and second biological samples, and therefrom monitoring the effectiveness of the therapy or immunization in the patient.

26. A method according to claim 25, wherein the step of detecting comprises (a) incubating the immunocomplexes with a detection reagent that is capable of binding to the immunocomplexes, wherein the detection reagent comprises a reporter group, (b) removing unbound detection reagent, and (c) detecting the presence or absence of the reporter group.

27. A method according to claim 26, wherein the detection reagent comprises a second antibody, or antigen-binding fragment thereof, capable of binding to the antibodies that specifically bind to the WT1 polypeptide.

28. A method according to claim 26, wherein the detection reagent comprises Protein A.

29. A method according to claim 26, wherein the reporter group is selected from the group consisting of radioisotopes, fluorescent groups, luminescent groups, enzymes, biotin and dye particles.

30. A method according to claim 25 wherein a reporter group is bound to the WT1 polypeptide, and wherein the step of detecting comprises removing unbound WT1 polypeptide and subsequently detecting the presence or absence of the reporter group.

31. A method for monitoring the effectiveness of an immunization or therapy for a malignant disease associated with WT1 expression in a patient, comprising the steps of:

(a) incubating a first biological sample with one or more of:

(i) a WT1 polypeptide;

(ii) a WT1 polynucleotide encoding a WT1 polypeptide; or

(iii) an antigen-presenting cell that expresses a WT1 polypeptide;

wherein the biological sample comprises CD4+ and/or CD8+ T cells and is obtained from a patient prior to a therapy or immunization, and wherein the incubation is performed under conditions and for a time sufficient to allow specific activation, proliferation and/or lysis of T cells;

(b) detecting an amount of activation, proliferation and/or lysis of the T cells;

(c) repeating steps (a) and (b) using a second biological sample comprising CD4+ and/or CD8+ T cells, wherein the second biological sample is obtained from the same patient following therapy or immunization; and

(d) comparing the amount of activation, proliferation and/or lysis of T cells in the first and second biological samples, and therefrom monitoring the effectiveness of the therapy or immunization in the patient.

32. A method according to claim 25 or claim 31, wherein the malignant disease is a cancer or a leukemia.

33. A method for determining the presence or absence of a malignant disease associated with WT1 expression in a patient, comprising the steps of:

(a) incubating $CD4^+$ T cells isolated from a patient with one or more of:

(i) a WT1 polypeptide;

(ii) a polynucleotide encoding a WT1 polypeptide; or

(iii) an antigen presenting cell that expresses a WT1 polypeptide; and

(b) detecting the presence or absence of specific activation of the T cells, therefrom determining the presence or absence of a malignant disease associated with WT1 expression.

34. A method according to claim 33, wherein the malignant disease is a cancer or a leukemia.

35. A method according to claim 33, wherein the step of detecting comprises detecting the presence or absence of proliferation of the T cells.

36. A method for determining the presence or absence of a malignant disease associated with WT1 expression in a patient, comprising the steps of:

(a) incubating $CD8^+$ T cells isolated from a patient with a one or more of:

(i) a WT1 polypeptide;

(ii) a polynucleotide encoding a WT1 polypeptide; or

(iii) an antigen presenting cell that expresses a WT1 polypeptide; and

(b) detecting the presence or absence of specific activation of the T cells, thereby determining the presence or absence of a malignant disease associated with WT1 expression.

37. A method according to claim 36, wherein the malignant disease is a cancer or a leukemia.

38. A method according to claim 36 wherein the step of detecting comprises detecting the presence or absence of generation of cytolytic activity.

39. A method for determining the presence or absence of a malignant disease associated with WT1 expression in a patient, comprising the steps of:

(a) incubating a biological sample obtained from a patient with one or more of:

(i) a WT1 polypeptide;

(ii) a polynucleotide encoding a WT1 polypeptide; or

(iii) an antigen presenting cell that expresses a WT1 polypeptide;

wherein the incubation is performed under conditions and for a time sufficient to allow immunocomplexes to form; and

(b) detecting immunocomplexes formed between the WT1 polypeptide and antibodies in the biological sample that specifically bind to the WT1 polypeptide; and therefrom determining the presence or absence of a malignant disease associated with WT1 expression.

40. A method according to claim 39, wherein the malignant disease is a cancer or a leukemia.

41. A method according to claim 39, wherein the step of detecting comprises (a) incubating the immunocomplexes with a detection reagent that is capable of binding to the immunocomplexes, wherein the detection reagent comprises a reporter group, (b) removing unbound detection reagent, and (c) detecting the presence or absence of the reporter group.

42. A method according to claim 41, wherein the detection reagent comprises a second antibody, or antigen-binding fragment thereof, capable of binding to the antibodies that specifically bind to the WT1 polypeptide.

43. A method according to claim 41, wherein the detection reagent comprises Protein A.

44. A method according to claim 41, wherein the reporter group is selected from the group consisting of radioisotopes, fluorescent groups, luminescent groups, enzymes, biotin and dye particles.

45. A method according to claim 39 wherein a reporter group is bound to the WT1 polypeptide, and wherein the step of detecting comprises removing unbound WT1 polypeptide and subsequently detecting the presence or absence of the reporter group.